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Changes of Purine Metabolism in Man and Animals Under Conditions of Molybdenum Biogeochemical Provinces

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CHANGES OF PURINE METABOLISM IN MAN AND ANIMALS UNDER CONDITIONS OF MOLYBDENUM BIOGEOCHEMICAL PROVINCES

/Izmeneniya purinovogo obmena u cheloveka i zhivotnykh v usloviyakh molibdenovykh biogeokhimicheskikh provintsiy/

by

V.V. Koval'skiy, G.A. Yarovaya and D.M. Shmavonyan,
V.I. Vernadskiy Institute of Geochemistry and Analytical Chemistry,
USSR Academy of Sciences, Moscow

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The links of the ecological chain from the change in chemical conditions of the external medium (a relative high molybdenum content in comparison to that of copper) to a change in purine metabolism in animal organisms and to the rise of an endemic gout-like sickness were followed up under the conditions of a molybdenum biogeochemical province. An increased molybdenum content in the soil leads to an increase of its content in plants and animals, and therefore in foodstuffs of people living in that region. Increased amounts of molybdenum entering the organism increased the activity of xanthine oxidase at the expense of an increase in its synthesis which, in its turn, leads to the formation of an augmented amount of uric acid in the organism.

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Sickness of animals with molybdenosis, caused by an excess molybdenum content in pasture plants, is encountered in England /11/, Canada /9/, USA /6/, New Zealand /8/ and other countries. This sickness is characterized by diarrhea, loss of weight and change in color and falling out of the fleece. The biochemical mechanisms of these manifestations of molybdenum toxicosis are still not clear.

After it became known that molybdenum is a part of the enzyme xanthine oxidase /27,21/, it could be assumed that the purine metabolism in an animal organism is disrupted in biogeochemical provinces enriched in molybdenum. With increase in molybdenum content of animal tissues, xanthine oxidase synthesis increases, which results in increased formation of uric acid in the organism and, under certain conditions, it can cause a gout type of sickness. No one has yet carried out research in this area.

To study the questions raised, biogeochemical provinces were selected with varying molybdenum content in the soil, feed and foodstuffs. In this case, the copper content in the environment was taken into account, since a connection of the functions of molybdenum and copper in metabolism in an animal organism is known /18,10/. An increase in the molybdenum content of the food causes a loss of copper from the organism. Addition of copper salts to the rations of animals and molybdenosis patients can remove the toxic effect of molybdenum on the body.

The research was conducted in the territories of two molybdenum biogeochemical provinces of Armenia, Kadzharana and Ankavana /5/. In the former, the molybdenum content was higher than in the reference

chernozem zone by fifty times in the soils on the average and 260 times in plants on the average, while it was 38 and 190 times, respectively, in the Ankavana province. However, the effect of molybdenum on animal organisms in Ankavana province was greater, since the copper content is relatively lower (Tables 1,2).

The increase in xanthine oxidase activity was considerably higher in the zone enriched with molybdenum than in the control and in the Ankavana province, compared with Kadzharana (Table 2).

The studies conducted enabled the question to be raised of the effect of the increased molybdenum concentration in the environment on human purine metabolism. This was of special interest, in connection with the customarily assumed absence of the enzyme uricase in his body. The Ankavana molybdenum biogeochemical province was selected for conduct of these studies, since, under its conditions, more significant changes in purine metabolism could have been expected, as a result of the excess molybdenum (with a relatively low copper content in the environment).

The tasks of the studies were:

1. Medical examination of the population of the molybdenum biogeochemical province, for the purpose of detection of gout-type illinesses;
2. Comparative determination of the amounts of molybdenum and copper entering the human body with food on the average, under the conditions of the Ankavana biogeochemical province, enriched with molybdenum, and under conditions of a control zone with a normal molybdenum content;
3. Study of the molybdenum and copper enrichment of the human body, by means of determination of the content of these elements in the blood

Table 1

Average molybdenum[1] and copper[2] content (in % of dry weight) in molybdenum province soils and plants

Объекта исследования (a)	b Каджаранская провинция			c Контрольная зона		
	Mo	Cu	Mo Cu	Mo	Cu	Mo/Cu
d Почвы	$1{,}0 \cdot 10^{-2}$ (57)	$9{,}8 \cdot 10^{-2}$ (57)	1 : 9,8	$4{,}7 \cdot 10^{-4}$ (37)	$5{,}2 \cdot 10^{-3}$ (37)	1 : 11,1
e Растения	$5{,}2 \cdot 10^{-3}$ (60)	$2{,}7 \cdot 10^{-2}$ (60)	1 : 5,2	$8{,}4 \cdot 10^{-5}$ (50)	$9{,}2 \cdot 10^{-4}$ (50)	1 : 11,0
	f Анкавинская провинция			c Контрольная зона		
d Почвы	$7{,}7 \cdot 10^{-3}$ (100)	$3{,}9 \cdot 10^{-3}$ (100)	2 : 1	$3{,}2 \cdot 10^{-4}$ (50)	$2{,}9 \cdot 10^{-3}$ (50)	1 : 9,1
e Растения	$3{,}8 \cdot 10^{-3}$ (210)	$8{,}5 \cdot 10^{-4}$ (210)	4,5 : 1	$6{,}1 \cdot 10^{-5}$ (70)	$5{,}8 \cdot 10^{-4}$ (70)	1 : 9,5

Key: a. Object of study
b. Kadzharana province
c. Control zone
d. Soil
e. Plants
f. Ankavana province

Note: The number of specimens analyzed is shown in parentheses.

/Translator's note: Commas in tabulated figures are equivalent to decimal points./

[1] Molybdenum determination was carried out by the thiocyanate method /1/.

[2] Copper determination was carried out, with the use of sodium diethyldithiocarbamate /4/.

Table 2

Xanthine oxidase[1] activity of sheep tissues in molybdenum biogeochemical provinces (in minutes on the average)

a Органы	b Каджаранская провинция			c Анкаванская провинция		
	d в зоне, обогащенной Mo	e в зоне, с нормальным содержанием Mo	f увеличение активности, %	d в зоне, обогащенной Mo	e в зоне с нормальным содержанием Mo	f увеличение активности, %
g Печень	2,80	3,76	34	2,28	4,69	106
h Почки	3,63	4,34	17	4,37	7,23	65
i Кишечник	6,14	8,40	37	4,40	11,88	170

Key:
a. Organ
b. Kadzharana province
c. Ankavana province
d. In Mo enriched zone
e. In zone with normal Mo content
f. Increase in activity, %
g. Liver
h. Kidneys
i. Intestines

[1] Xanthine oxidase determination was carried out under anaerobic conditions, with the use of methylene blue as the hydrogen accentuator /13,23/.

and urine with an increased molybdenum content in the food;

4. Study of the uric acid content in the blood and of its elimination with the urine;

5. Determination of blood xanthine oxidase activity;

6. Determination of the nature of the correlation of the increased molybdenum content in the food ration with the uric acid content in the food and its elimination from the body and the blood xanthine oxidase activity.

We studied the population of two populated places located in a molybdenum biogeochemical province, Ankavana village (former Miskhana), which has existed for over a hundred years, and a new settlement, which developed adjacent to it only six years ago.

Three hundred persons from Ankavan village underwent medical examination. Of them, 184 persons were age 18 and older, and 100 persons (78 adults) were from the new settlement. It was determined as a result of the examination that the characteristic clinical symptoms of gout are observed in a significant number of the inhabitants. Thus, in Ankavan village, gout symptoms were detected in 57 persons, which is 31% of the adult population and, in the new settlement in 14 persons (17.9%).

These figures are extremely high, since, according to statistical data, no more than 1-4% of the people are sick with gout on the average.

The illness diagnosed as gout, observed in the molybdenum biogeochemical province, is characterized by pains in the joints, especially in the knee and metatarsophalangeal joints of the legs and the phalangeal joints of the hands. Cases of well-expressed deformation of the joints are observed, as well as of swelling, elevated temperatures and reddening

Table 3

Molybdenum and copper content of human foodstuffs and drinking water (in % in dry matter in vegetable products, in fresh matter in animal)

a Продукты, из которых взяты пробы	b Район, обогащенный Mo		c Район с нормальным содержанием Mo	
	Mo	Cu	Mo	Cu
d Растительные продукты				
e Картофель	$1,1 \cdot 10^{-3}$	$3,2 \cdot 10^{-4}$	$3,3 \cdot 10^{-4}$	$7,0 \cdot 10^{-4}$
f Капуста	$5,2 \cdot 10^{-4}$	$5,9 \cdot 10^{-4}$	$0,4 \cdot 10^{-5}$	$2,2 \cdot 10^{-3}$
g Салат (котим)	$7,3 \cdot 10^{-4}$	$5,0 \cdot 10^{-4}$	$1,8 \cdot 10^{-4}$	$4,8 \cdot 10^{-3}$
h Помидоры	$8,5 \cdot 10^{-4}$	$2,9 \cdot 10^{-3}$	$2,7 \cdot 10^{-4}$	$1,7 \cdot 10^{-3}$
i Баклажаны	$1,3 \cdot 10^{-3}$	$8,7 \cdot 10^{-4}$	$9,7 \cdot 10^{-5}$	$9,3 \cdot 10^{-4}$
j Свекла	$1,8 \cdot 10^{-3}$	$6,5 \cdot 10^{-4}$	$3,2 \cdot 10^{-4}$	$2,1 \cdot 10^{-4}$
k Фасоль (лоби)	$8,2 \cdot 10^{-3}$	$1,7 \cdot 10^{-3}$	$5,1 \cdot 10^{-4}$	$1,4 \cdot 10^{-3}$
l Прангос (похи)	$2,1 \cdot 10^{-3}$	$4,4 \cdot 10^{-4}$	$9,0 \cdot 10^{-5}$	$5,1 \cdot 10^{-4}$
m Щавель (авелук)	$3,0 \cdot 10^{-4}$	$1,8 \cdot 10^{-4}$	$2,9 \cdot 10^{-5}$	$3,3 \cdot 10^{-4}$
n Мята	$5,7 \cdot 10^{-3}$	$4,8 \cdot 10^{-4}$	$1,1 \cdot 10^{-4}$	$7,7 \cdot 10^{-4}$
o Базилик (реан)	$5,9 \cdot 10^{-4}$	$9,7 \cdot 10^{-4}$	$4,6 \cdot 10^{-5}$	$9,0 \cdot 10^{-4}$
p Резак (сибех)	$9,3 \cdot 10^{-4}$	$1,0 \cdot 10^{-3}$	$7,3 \cdot 10^{-5}$	$1,5 \cdot 10^{-3}$
q Эстрагон (тархун)	$8,3 \cdot 10^{-4}$	$6,1 \cdot 10^{-4}$	$2,3 \cdot 10^{-5}$	$3,0 \cdot 10^{-4}$
r Лук порей (савзи)	$1,0 \cdot 10^{-3}$	$2,7 \cdot 10^{-3}$	$9,7 \cdot 10^{-5}$	$1,2 \cdot 10^{-3}$
s Хлеб белый из привозной муки	$5,1 \cdot 10^{-5}$	$2,1 \cdot 10^{-4}$	$3,9 \cdot 10^{-5}$	$2,8 \cdot 10^{-4}$
t Хлеб черный из привозной муки	$2,8 \cdot 10^{-5}$	$7,4 \cdot 10^{-5}$	$4,1 \cdot 10^{-5}$	$5,9 \cdot 10^{-4}$
u Животные продукты				
v Молоко коровье	$2,8 \cdot 10^{-5}$	$1,9 \cdot 10^{-5}$	$4,7 \cdot 10^{-6}$	$2,9 \cdot 10^{-5}$
w Печень говяжья	$3,4 \cdot 10^{-5}$	$2,1 \cdot 10^{-4}$	$9,8 \cdot 10^{-6}$	$1,1 \cdot 10^{-3}$
x Печень баранья	$2,4 \cdot 10^{-5}$	$1,7 \cdot 10^{-5}$	$8,6 \cdot 10^{-6}$	$7,4 \cdot 10^{-4}$
y Мясо говяжье	$5,9 \cdot 10^{-6}$	$4,0 \cdot 10^{-5}$	$1,2 \cdot 10^{-6}$	$8,2 \cdot 10^{-5}$
z Мясо баранье	$3,4 \cdot 10^{-6}$	$5,1 \cdot 10^{-5}$	$1,0 \cdot 10^{-6}$	$6,8 \cdot 10^{-5}$
a' Яйца	$8,1 \cdot 10^{-7}$	$4,9 \cdot 10^{-5}$	$3,4 \cdot 10^{-7}$	$9,1 \cdot 10^{-5}$
b' Вода питьевая	$1,0 \cdot 10^{-6}$	$2,7 \cdot 10^{-5}$	$4,7 \cdot 10^{-7}$	$1,1 \cdot 10^{-5}$

Note: Local names are given in parentheses.

Key:

a. Products sampled
b. Region enriched with Mo
c. Region with normal Mo content
d. Vegetable products
e. Potato
f. Cabbage
g. Lettuce (kotim)
h. Tomatoes
i. Eggplants
j. Beets
k. Beans (lobi)
l. Prangos (pokhi)
m. Dock (aveluk)
n. Mint
o. Basil (rean)
p. Sedge (sibekh)
q. Tarragon (tarkhun)
r. Leek (savzi)
s. White bread from imported flour
t. Black bread from imported flour
u. Animal products
v. Cow's milk
w. Beef liver
x. Sheep liver
y. Beef
z. Mutton
a'. Eggs
b'. Drinking water

in the region of the joints. Acute attacks with increased temperatures are rarely observed. In all cases, the illness is accompanied by an increase in uric acid content in the blood.

In a significant number of cases, the gout illness of the people under molybdenum province conditions was accompanied by illnesses of the gastrointestinal tract, liver, and kidneys (positive Pasternatskiy reaction). Enlargement of the liver was observed in the majority of people in whom symptoms of gout were found.

Fifty-two persons among the local inhabitants and five persons living outside the molybdenum province (for the control) underwent detailed examination. Determination of the molybdenum, copper, uric acid and blood xanthine oxidase activity, as well as daily elimination of molybdenum, copper and uric acid with the urine, were carried out.

Before proceeding to description of these studies, the results of determination of the molybdenum and copper in the foodstuffs and rations of the people living in the molybdenum province must be reported.

The population of Ankavan village and of the new settlement eat mainly products of local origin. We examined the basic foodstuffs of the people living in these populated places for their molybdenum and copper content and, for comparison, such studies were carried out in control regions located outside the molybdenum province. The analytical results are presented in Table 3.

It has been determined that the foodstuffs of the population[1]

[1]Average composition of daily (summer) ration, calculated on the basis of questioning 21 persons: rye and wheat bread, 500 g; wheat flour, 30 g; grits and macaroni, 50 g; potatoes, 400 g; fresh vegetables (cabbage, eggplant, tomatoes, beets etc.), 300 g; beans, 100 g; greens, 100 g; sugar, 50 g; meat and meat products, 200 g; milk and milk products, 600 g; eggs, 1; water, 1200 g.

of the molybdenum province contain 2-10 times more than the products from control areas. According to information obtained from the local population, products of local origin make up at least 50% of the total ration of an adult person. On the basis of the analyses carried out and of questioning the population, we calculated that, on the average, an adult person in the zone of the Ankavana molybdenum province receives 10-15 mg of molybdenum and 5-10 mg of copper daily with food, while normally outside the molybdenum province a person receives 1-2 mg of molybdenum and 10-15 mg of copper with food.

The high molybdenum content, with a relatively low copper content, in the daily human food ration affects concentration of molybdenum by the body to a definite extent. This is reflected by the molybdenum and copper content of the blood and elimination with the urine (Table 4). We observed an increased molybdenum content in the blood in nearly all the inhabitants of the Ankavana biogeochemical province with some decrease in copper concentration compared with the control (outside the molybdenum province). As a rule, the daily quantity of urine contains an increased amount of molybdenum and copper, compared with the amount of them in the urine of people of the control regions.

Summary results of biometric processing of the data obtained are presented in Tables 4-7. The value of t permits a decision as to how confidently M_1, M_2, etc. can be considered, not as variants of the same M, but as belonging to different variation series. It has been proved theoretically /3,2/ that, if one series is subtracted from another

Table 4

Molybdenum and copper content of human blood and urine

a Район	d Объект исследования	e Число случаев	f Представительные величины	a Кровь: g Содержание Mo (мг %)	a Кровь: h Содержание Cu (мг %)	a Кровь: Cu/Mo	a Кровь: e Число случаев	b Моча: i Содержание Mo (мг в суточном количестве мочи)	b Моча: j Содержание Cu (мг в суточном количестве мочи)	b Моча: Cu/Mo
Молибденовая провинция	Больные k	16	M_1^*	0,031	0,113	3,5	6	0,079	0,304	10,5
			$\pm s_1^*$	0,008	0,025	2,7		0,002	0,014	2,2
			$\pm m_1^*$	0,002	0,006	0,68		0,001	0,006	0,55
	Здоровые l	31	M_2	0,017	0,138	10,2	11	0,028	0,185	7,2
			$\pm s_2$	0,008	0,026	4,8		0,005	0,030	3,9
			$\pm m_2$	0,001	0,005	0,86		0,001	0,009	0,97
			t_{1-2}^*	6,4	3,1	5,2		0,7	12,0	2,7
	Среднее в провинции (больные + здоровые) m	47	M_3	0,022	0,129	8,2	17	0,028	0,226	8,4
			$\pm s_3$	0,010	0,030	2,1		0,003	0,032	0,3
			$\pm m_3$	0,001	0,004	0,30		0,001	0,008	0,07
Контрольная зона o	Контрольные p	5	M_4	0,006	0,183	33,7	4	0,016	0,120	7,2
			$\pm s_4$	0,002	0,020	8,9		0,003	0,024	1,3
			$\pm m_4$	0,001	0,009	4,00		0,002	0,012	0,59
			t_{2-4}	7,1	4,5	5,7		5,5	4,6	0
			t_{3-4}	11,4	5,5	6,4		5,5	7,6	0,3

Note: M is the arithmetic mean; $\pm$ m is the mean error $\left(m = \frac{\sigma}{\sqrt{n}}\right)$; $\pm\sigma$ is the root mean deviation $\left(\sigma = \sqrt{\frac{\sum p \cdot a^2}{n-1}}\right)$, where p is the number of variants, a is the deviation from the mean, n is the number of cases); $t = \frac{M_1 - M_2}{\sqrt{m_1^2 + m_2^2}}$.

Key:
a. Blood
b. Urine
c. Region
d. Object of study
e. Number of cases
f. Representative quantities
g. Mo content (mg %)
h. Cu content (mg %)
i. Mo content (mg in daily amount)
j. Cu content (mg in daily amount)

Key: k. Sick persons
l. Healthy persons
m. Average in province (sick + healthy)
n. Molybdenum province
o. Control zone
p. Controls

which is being compared with it and the difference is then divided into the square root of the sum of the squares of the errors of these arithmetic means, in the event the difference is random, the resulting partial will be 2-3 units less. If the partial is greater, this means that neither value can be a partial reflection of one general series. The larger the value of t, the more significant the difference between the variation series being compared. Thus, for example, in comparing the molybdenum content of the blood of people, gout patients and healthy ones, living under the molybdenum province conditions, despite the appreciable transgression of one series on the other, they differ quite strongly from each other ($t = 6.4$); this difference is still more appreciable, in comparison of the blood molybdenum content of people living in the province and controls living outside the enrichment zone; in this case, the difference of the means exceeds its error by 11.4 times.

The dependence of blood copper content and its elimination with urine on blood molybdenum content is shown in Fig. 1. It can be seen there that the increase in amount of molybdenum in the blood results in some decrease in its copper content and to considerable increase in elimination of the latter with the urine. The curve depicted in Fig. 2 shows that, with decrease in the blood Cu/Mo ratio, the value of this ratio for the urine increases. Data obtained on people confirm the known fact of antagonism between copper and molybdenum in an animal organism. Increase in food and, hence, tissue molybdenum content leads to more intensive elimination of

copper from the organism and a decrease in reserves of it in the tissues and, moreover, it indicates a possible accumulation of molybdenum in the human body, under conditions of an increased content of it in the environment.

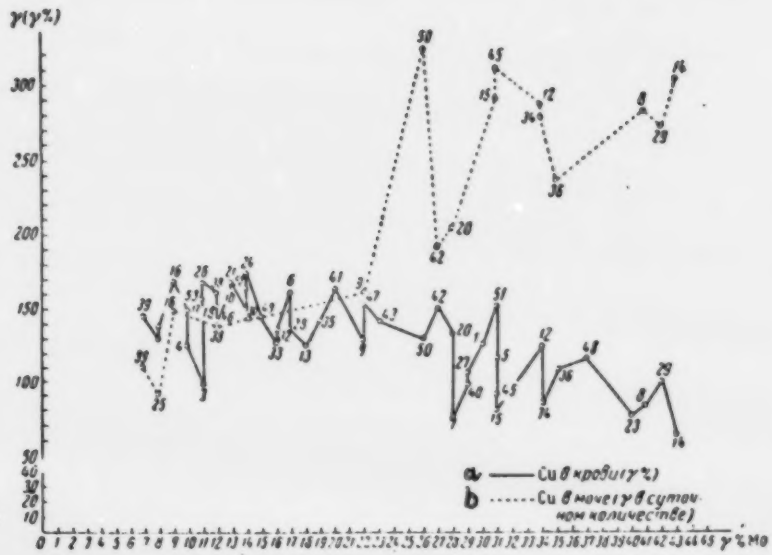

Fig. 1. Copper content of blood and its elimination with the urine vs. molybdenum content of the blood (the figures on the curves correspond to the number of cases).

Key: a. Cu in blood (γ %)
b. Cu in urine (γ in daily amount)

This gives a basis for the proposal that excess molybdenum can be partially consumed by synthesis of additional amounts of xanthine oxidase, the molecule of which always contains this metal. The increase in the amount of xanthine oxidase in the blood can be decided in this case, by determination of the enzyme activity.

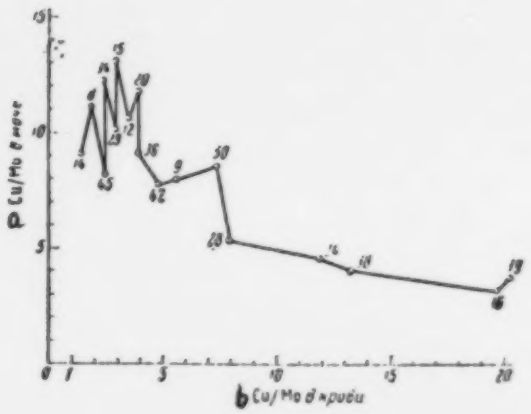

Fig. 2. Change in Cu/Mo ratio in the urine vs. value of this ratio in the blood.

Key: a. Cu/Mo in urine
b. Cu/Mo in blood

To test our hypothesis of a possible connection between xanthine oxidase activity and the amount of molybdenum received with food by people living in the area of a province with an increased molybdenum content, human blood xanthine oxidase activity was determined. Since the method which we adapted for determination of the activity of this enzyme under field conditions is based on the methylene blue bleaching reaction, it did not seem possible to use whole blood to measure the activity. Therefore, considering indications in the literature of the occurrence of xanthine oxidase in blood serum /26/, we measured the activity of the enzyme in human blood serum. The determination was carried out 30 min after taking blood from a vein (during this time,

the blood was kept at a temperature of +3°).

Xanthine oxidase activity was measured in 19 persons from the molybdenum province and in five persons of the controls.

The test results are presented in Table 5.

Table 5

Xanthine oxidase activity in human blood serum

a Район	b Объект исследования	c Представленная величина	d Число обследованных	e Активность в минутах
f Молибденовая провинция	Больные g	M_1 $\pm\sigma_1$ $\pm m_1$	8	4,77 0,53 0,189
	Здоровые h	M_2 $\pm\sigma_2$ $\pm m_2$ t_{1-2}	11	7,89 0,97 0,293 8,9
	Среднее в провинции i	M_3 $\pm\sigma_3$ $\pm m_3$	19	6,59 2,17 0,491
Контрольная зона j	Контрольные k	M_4 $\pm\sigma_4$ $\pm m_4$ t_{2-4} t_{3-4}	34	10,19 1,41 0,641 3,3 4,5

Key:
a. Region
b. Object of study
c. Quantity
d. Number of cases
e. Activity in minutes
f. Molybdenum province
g. Sick persons
h. Healthy persons
i. Average in province
j. Control zone
k. Controls

As is seen from Table 5, an increase in activity of the enzyme takes place in parallel with the increase in blood molybdenum content (Table 4). This gives a basis for considering that an increase in amount of molybdenum in an organism (within the limits of molybdenum concentrations which we found) results in an increase in xanthine oxidase synthesis.

This dependence of xanthine oxidase activity on molybdenum content of the blood, as well as on the Cu/Mo ratio, is presented graphically in Figs. 3 and 4, where an increase in xanthine oxidase activity with increase in Mo content and a decrease in the Cu/Mo ratio is seen. The jumps observed in the curves can evidently be explained by the individual characteristics of metabolism of various people.

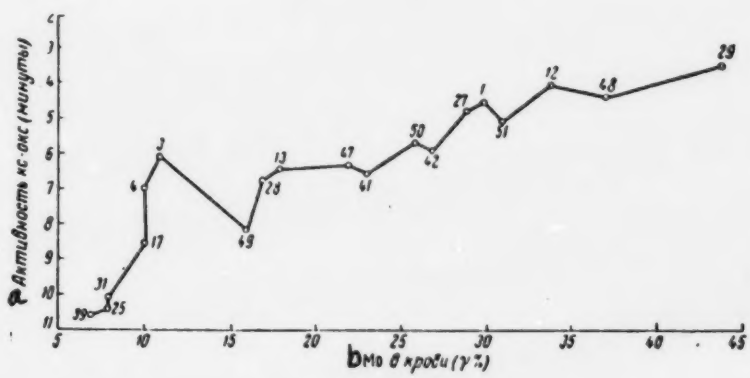

Fig. 3. Xanthine oxidase activity vs. molybdenum content of blood.

Key: a. Xanthine oxidase activity (minutes)
b. Mo in blood (γ %)

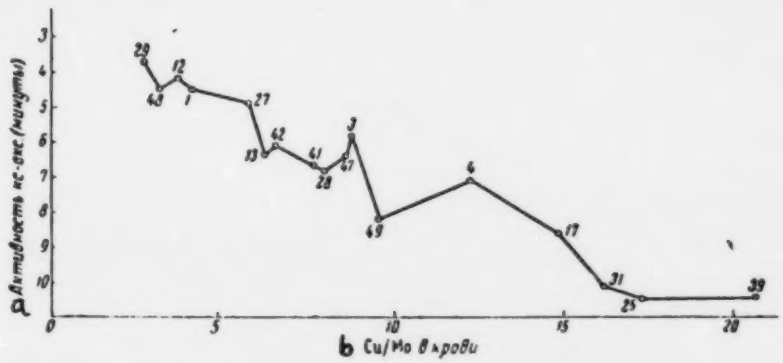

Fig. 4. Xanthine oxidase activity vs. Cu/Mo ratio of the blood.

Key: a. Xanthine oxidase activity (minutes)
b. Cu/Mo in blood

The correlation established between the increase in molybdenum content of the blood, reflecting an accumulation of molybdenum in the body, and the increased xanthine oxidase activity is very important for understanding the processes of purine metabolism in the molybdeum province.

The increased xanthine oxidase activity should promote an increase in formation of uric acid in the body.

In fact, such a relationship was successfully demonstrated. In comparing the data of blood uric acid content[1] (Table 6) and blood xanthine oxidase activity (Table 5), a direct dependence can be observed; the uric acid concentration increases together with the increase in xanthine oxidase activity (Fig. 5).

[1]Uric acid was determined with phosphotungstate reagent /12,15/.

Table 6

Uric acid content of human blood and urine

a Район	b Объект исследования	c Число случаев	d Предст. величины	e Кровь: g Содержание мочевой кислоты (мг %)	e Кровь: c Число случаев	f Моча: h Содержание мочевой кислоты (мг в сутки)
i Молибденовая провинция	j Больные	17	M_1	8,1	17	823,8
			$\pm\sigma_1$	1,77		284,8
			$\pm m_1$	0,43		67,2
	k Здоровые	35	M_2	5,3	34	648,7
			$\pm\sigma_2$	2,09		233,5
			$\pm m_2$	0,35		39,6
			t_{1-2}	5,1		2,2
	l Среднее в провинции (больные + здоровые)	52	M_3	6,2	51	707,0
			$\pm\sigma_3$	2,3		246,8
			$\pm m_3$	0,32		42,5
m Контрольная зона	n Контрольные	5	M_4	3,8	5	432,1
			$\pm\sigma_4$	1,01		88,6
			$\pm m_4$	0,46		40,3
			t_{2-4}	2,6		3,9
			t_{3-4}	4,4		4,7

Key:
a. Region
b. Object of study
c. Number of cases
d. Quantity
e. Blood
f. Urine
g. Uric acid content (mg %)
i. Molybdenum province
j. Sick persons
k. Healthy persons
l. Average in province (sick + healthy)
m. Control zone
n. Controls

Changes in purine metabolism taking place in specific body tissues evidently are reflected in the blood in this case.

The well-expressed relationship of uric acid and molybdenum

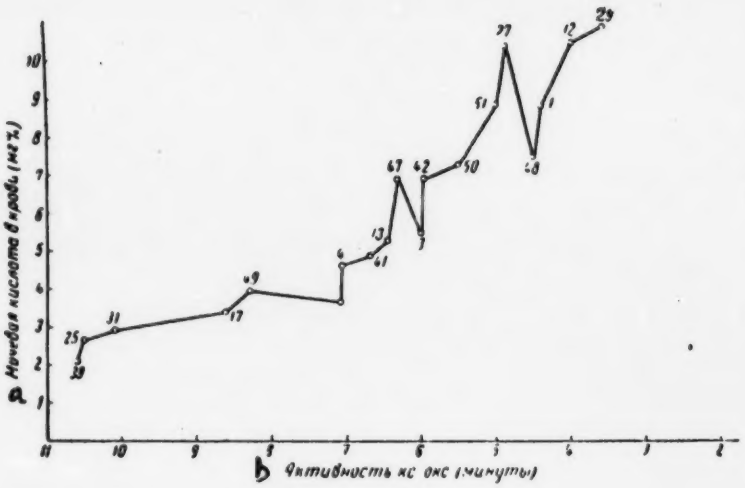

Fig. 5. Blood uric acid content vs. xanthine oxidase activity.

Key: a. Blood uric acid content (mg %)
b. Xanthine oxidase activity (minutes)

content of the blood is completely natural under these conditions (Figs. 6 and 7).

The significant distribution of gout type diseases among people in the molybdenum biogeochemical province becomes of great interest, in the light of the material presented on molybdenum and copper metabolism, xanthine oxidase activity and accumulation of uric acid in the body.

In the overwhelming majority of cases, the presence of gout type

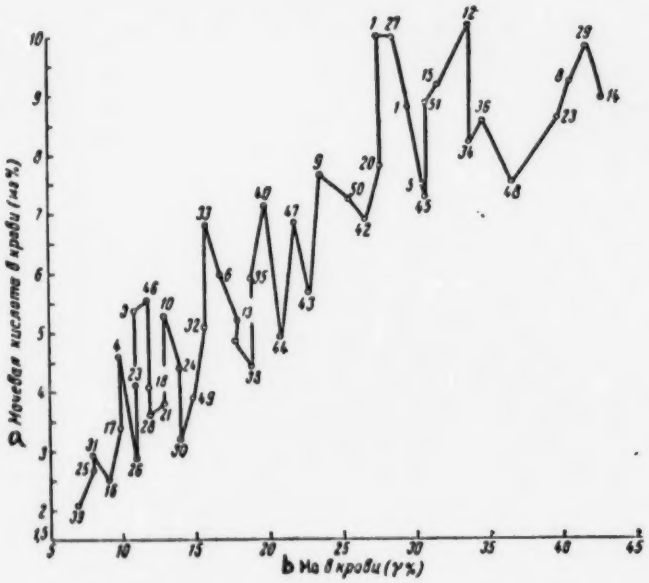

Fig. 6. Uric acid content vs. amount of molybdenum in blood.

Key: a. Blood uric acid (mg %)
b. Mo in blood (γ %)

disease symptoms coincided with an increase in th. blood uric acid content. In the survey, gout symptoms were recorded in 18 of 52 persons, which is 34.6%.

An above normal uric acid content of the blood (over 5.5 mg %) /16,30/ was found in 29 persons of 52 examined, i.e., in 55.7%. Of them, gout symptoms were found in a preliminary examination in 17 persons, which included deformation of the joints in five cases,

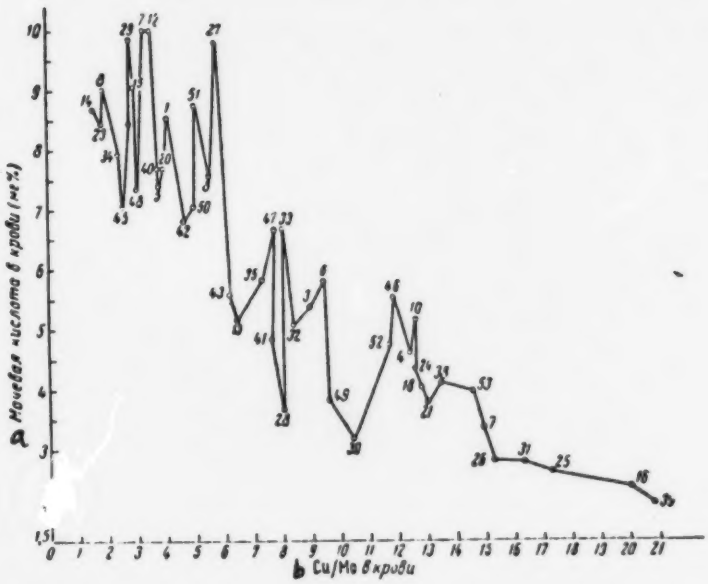

Fig. 7. Blood uric acid content vs. Cu/Mo ratio.

Key: a. Blood uric acid (mg %)
b. Cu/Mo in blood

sufficient symptoms were not found for a more or less firm conclusion in six persons and six persons had no clinical symptoms of gout.

A dependence of blood uric acid content (on the average) on lifetime under Ankavana province conditions was established: with a stay time of up to one year, 3.75 mg %, those living from one year to five years, 6.4 mg % and in persons in this region five years or more, 6.8 mg %. Obviously, the changes developed quite quickly, during the first year

of life in this region.

Elimination of uric acid with the urine by the persons examined remained basically within normal limits /19/ and, as a rule, it increased somewhat with increase in blood uric acid concentration. A sharp decrease in uric acid elimination, observed with a high content of it in the blood, was noted in five cases. Renal insufficiency was established in three of them, and the other two had the chronic form of gout, i.e., evidently the low elimination of uric acid from the body was caused by kidney disease, which in all likelihood is secondary in this case. Quite frequent kidney diseases of the inhabitants of Ankavana, coinciding with the presence of gout, can indicate a secondary kidney insufficiency developing in gout.

In animals, molybdenum toxicosis, accompanied by an increased elimination of copper from the organism, is cured by addition of copper to the food. In this case, elimination of molybdenum from the body increases. It is completely likely that, in people in whom we observed an increase in elimination of copper with increase in blood Mo content, small additions of copper to the food might increase elimination of molybdenum from the body and thereby prevent the disease. So much the more that, in regions of other molybdenum provinces (the Kadzharana and Dastakert regions), where, together with a high Mo content in the environment, there is a still higher amount of copper, 10-15 times more than the molybdenum content (Tables 1 and 2), no signs of a disease similar to gout are observed. In all likelihood, this is a result of the high copper content in the food. Data from the determination of the uric acid content in the blood, its elimination with the urine, molybdenum and copper concentrations in the

blood and their elimination of the inhabitants of Kadzharana are presented in Table 7.

Table 7

Content of molybdenum, copper and uric acid in the blood and their elimination with the urine of people living in the Kadzharana molybdenum province.

a Число случаев	b Предст. величины	c Кровь: e Содержание Mo (мг %)	c Кровь: f Содержание Cu (мг %)	c Кровь: g Содержание мочевой кислоты (мг %)	d Моча: h Содержание Mo (мг в сутки)	d Моча: i Содержание Cu (мг в сутки)	d Моча: j Содержание мочевой кислоты (мг в сутки)
15	M_b	0,010	0,213	3,4	0,068	0,463	582,8
	σ_b	0,003	0,030	0,837	0,013	0,077	181,0
	m_b	0,001	0,010	0,218	0,005	0,027	46,9
	t_{1-5}	9,6	8,3	21,3	7,8	4,8	3,0
	t_{2-5}	5,0	6,8	8,7	8,0	9,1	1,1
	t_{4-5}	4,0	2,3	1,8	10,4	10,5	3,4

Key:
a. Number of cases
b. Quantity
c. Blood
d. Urine
e. Mo content (mg %)
f. Cu content (mg %)
g. Uric acid content (mg %)
h. Mo content (mg per day)
i. Cu content (mg per day)
j. Uric acid (mg per day)

DISCUSSION OF RESULTS

To a certain extent, we succeeded in following the links of the ecological chain leading from change in the biogeochemical conditions of the environment (relatively high molybdenum content compared to

copper) to changes in the purine metabolism in an animal organism and the development of endemic gout-type illinesses. An increased molybdenum content in the soil results in an increase in the amount of this element in plant and animal organisms and, consequently, in the foodstuffs of people living in this region. Upon entry of increased amounts of molybdenum into the organism,[1] the xanthine oxidase activity increases, owing to an increase in synthesis of it. In turn, this results in formation of an increased amount of uric acid in the organism.

We have not observed a noticeable increase of uric acid in animal tissues. This evidently is explained by the presence of active uricase in the animals. Upon entry of considerable amounts of molybdenum into the organism, the difference in human and animal purine metabolism is detected by accumulation of considerable amounts of uric acid in the human body.

We consider that our hypothesis that the illness observed in the molybdenum province of Ankarana is a gout-type illness is based, to a considerable extent, on the observed clinical picture of the disease, similar to clinically known cases of gout /17,24/.

Moreover, we have observed good coincidence of the preliminary clinical diagnosis of "gout" with a high uric acid content in the blood. The fact that the clearly expressed gout nodes are observed only in some persons does not contradict the conclusion as to the nature of the illness, since accumulation of urates is frequently noted in gout

[1] A still higher Mo content in the food can cause another effect and even decrease xanthine oxidase activity (the data were obtained on rats, in the biogeochemical laboratory of N. Morozova).

without tophi /28/, especially in the initial stage of it.

An increase in uric acid concentration in the blood, together with the increase in elimination of it observed in our studies, is in agreement with the opinions of those authors who point out that in the majority of cases, especially at the start of gout, the kidneys remove uric acid well /7,14,25/. A decrease in uric acid excretion usually is secondary in gout patients.

The most widespread theory concerning the origin of gout is the theory of increased uric acid synthesis /19/. The question of the nature of the increase of uric acid concentration in gout still remains open. Gutman /14/ thinks gout causes disorders in the early processes of purine synthesis (for example, nucleoside phosphorylation). Others think /29/ that, in gout, disruption of nucleotide synthesis from simple precursors occurs (rapid conversion of excess purine bases to uric acid occurs after excessive purine synthesis). However, the question of the importance of the increase in xanthine oxidase activity as an etiological factor in gout still has not been examined in known literature.

On the basis of what has been presented, we consider an increased molybdenum content in the food to be a possible cause of gout, depending on geochemical factors: the molybdenum content and mobility in the soil. The significant distribution of gout in the territory of a biochemical province enriched with molybdenum permits it to be considered that the existence of endemic gout or of endemic gout-type illnesses is possible.

It can be considered that an increase in uric acid synthesis takes place, owing to an increase in xanthine oxidase activity, the more so

that the final formation of uric acid always takes place in a stage catalyzed by xanthine oxidase. In other words, no matter how synthesis of the uric acid nucleus took place before formation of hypoxanthine, direct formation of uric acid always takes place only with the assistance of xanthine oxidase /22/. Xanthine oxidase is an enzyme which contains molybdenum. Its synthesis depends on the molybdenum content in the organism, which in turn is determined by the molybdenum content of the geochemical environment.

Our data concerning possible participation of molybdenum in the etiology of gout, on condition of a high content of it in food, only point out still another of the possible causes of development of this disease not noted in the literature and, to a certain extent, it reinforces the role of an increase in uric acid synthesis in the development of gout.

CONCLUSIONS

Under the conditions of a molybdenum biogeochemical province, purine metabolism of people is disrupted, and an endemic illness develops, with basic symptoms of gout. In this case, an increase in molybdenum content in the body and an increased elimination of copper, an increase in xanthine oxidase activity and an increase in uric acid concentration in the blood are observed.

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